

Detection of Antibody to Group B Adult Diarrhea Rotaviruses in Humans

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Group B rotaviruses have been responsible for annual epidemics of severe diarrhea affecting both adults and children in China. We developed a specific and sensitive enzyme-linked immunosorbent blocking assay to detect antibody to group B rotaviruses that will be useful to assess the role of group B rotavirus infections as a cause of human gastroenteritis. We tested 219 human sera and 18 immunoglobulin pools collected from eight countries for antibodies to both group A and group B rotaviruses. Overall, a low proportion (10 of 237 or 4.2%) of sera contained antibody to group B rotaviruses. Antibody to group B rotavirus was detected in only 1 of 155 serum samples from healthy or hospitalized individuals in the United States, including patients with the chronic inflammatory bowel diseases Crohn's disease and ulcerative colitis. No antibody was detected in 15 serum samples from Australia and from an outbreak of gastroenteritis on a cruise ship or in nine immunoglobulin pools from Japan and the United Kingdom. Antibody to group B rotaviruses was detected in 8 convalescent- (but not acute-) phase serum samples from Chinese patients with group B gastroenteritis, in five immunoglobulin pools from China, in 1 of 6 serum samples from Chinese students in the United States, and in 1 each of 10 serum samples from Kenya, 20 from Thailand, and 15 from Canada. In contrast, most of these samples (226 of 237 or 95.4%) had antibody to group A rotaviruses. These results indicate that human infection with group B rotavirus has not been widespread in areas outside China. Seroconversion observed between the acute- and convalescent-phase serum samples from China also suggests that infections with this virus are primary infections. Continued surveillance for this new group of rotaviruses should determine whether the many susceptible people become infected or whether other factors influence the severe pathogenicity of human infections with these viruses in China.

Rotaviruses are proposed to be subdivided into groups, with viruses in each group sharing cross-reactive antigens located on one or more of the capsid proteins (11). Five rotavirus groups (to be designated A, B, C, D, and E) have been proposed based on comparative antigenic and genetic studies primarily with animal rotaviruses (22-24). Similar studies on the non-group A adult diarrhea rotaviruses (ADRV), which have caused annual epidemics of diarrhea among both adults and children in China (17, 29), have shown that these viruses should be classified as group B rotaviruses (7, 21). Infections with the ADRV have been extremely severe (choleralike) in all age groups, and mortality resulting from severe dehydration was reported in some elderly patients in China (25). Preliminary seroepidemiologic studies with sera from areas (Hong Kong and Australia) where ADRV have not been identified reported a low prevalence of antibody to ADRV (16).

Information on the prevalence of antibody to the ADRV in human sera from the United States has not been reported. However, a high prevalence of antibody to a non-group A rotaviruslike agent was reported in healthy adults and children in Baltimore (9). Symptomatic gastroenteritis in adults and children caused by this virus has also been reported in Baltimore (9, 28). This non-group A rotavirus has been unclassified, but our present studies indicate it is a group B rotavirus.

We recently developed an enzyme-linked immunosorbent assay (ELISA) to detect group B rotaviruses (21). We now

report the extension of this antigen ELISA to a blocking ELISA (ELISA-BL) to detect antibody to group B rotaviruses. We used this ELISA-BL to evaluate the prevalence of antibody to the ADRV in human sera from different areas of the world.

MATERIALS AND METHODS

Serum specimens. A total of 237 serum samples and 18 immunoglobulin pools were tested by ELISA-BL for antibody to group B ADRV. Nine paired serum samples from patients with gastroenteritis caused by ADRV were from an outbreak in Nanning, Guangxi, People's Republic of China (29). Nineteen serum samples were collected from staff members of the Department of Virology, Baylor College of Medicine, Houston, Tex., in 1985 and 1986; six of those samples were from Chinese staff who came from the People's Republic of China after 1983, and three samples were from Chinese or Japanese staff from Hong Kong, Taiwan, or Japan. Thirty serum samples were obtained from 10 healthy adults in Houston and 20 patients with chronic idiopathic inflammatory bowel disease (Crohn's disease or ulcerative colitis) in Houston and New York, N.Y. Fifty-seven serum samples were collected from children, ages 1 month to 19 years, who were admitted to Texas Children's Hospital without gastroenteritis in 1986. Sera from Thailand, Kenya, and Australia were collected between 1981 and 1985 from healthy adults as part of a study of neoplasia and steroid contraceptives. Thirty-five paired serum samples from adults with gastroenteritis of unknown origin (from the United States, Canada, and a cruise ship) were from the

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collection of the Viral Gastroenteritis Laboratory of the Centers for Disease Control, Atlanta, Ga. Twenty-five serum samples were from patients admitted to Ben Taub General Hospital in Houston in 1986 for unknown illnesses. Thirteen samples from patients without gastroenteritis were supplied by Sudhic Dutta (Veterans Administration Medical Center, Baltimore, Md.). Thirteen commercial immunoglobulin pools from adults from the United Kingdom, Canada, and Japan (D. Cubitt and D. McSwiggan, *J. Med. Virol.*, in press) were kindly supplied by D. Cubitt (Public Health Laboratory Service, Central Middlesex Hospital, London, United Kingdom). Five immunoglobulin pools each from 2,000 adult donors from China were kindly supplied by Jian-Zi Xiang (Shanghai Institute of Biological Products, Ministry of Public Health, Shanghai, People's Republic of China).

Hyperimmune antisera. Hyperimmune guinea pig antiserum to SA11, hyperimmune guinea pig and rabbit antiserum to group B ADRV, and hyperimmune guinea pig antiserum to the porcine group C rotavirus (Cowden strain) were prepared as described previously (12, 21). Mouse hyperimmune antiserum to group B ADRV was prepared by inoculating animals with virus purified from stools as previously described (19). Guinea pig and mouse hyperimmune antisera to group B ADRV, hyperimmune gnotobiotic pig antiserum to porcine group B rotavirus, gnotobiotic calf antiserum to bovine group B rotavirus, gnotobiotic pig antiserum to porcine group C rotavirus, and chicken and rat antisera to the non-group A rat rotavirus were provided by T. Hung (Institute of Virology, Beijing, People's Republic of China), L. Saif (Ohio State University, Wooster), and R. Yolken (Johns Hopkins University School of Medicine, Baltimore, Md.) and were prepared and characterized as previously described (3, 16, 21, 27, 28).

ELISA-BL for antibody to rotaviruses. The ELISA-BL to detect antibody was developed by modifying our previously described ELISA for group B antigen(s) (21). Phosphate-buffered saline (PBS; pH 7.4) containing 10% normal guinea pig serum and 1% bovine serum albumin was used as diluent for antigen and for the peroxidase-conjugated goat antibody to rabbit immunoglobulin G. Briefly, the antigen was prepared from a single stool specimen derived from an outbreak of group B ADRV gastroenteritis (29) using extraction with trichlorotrifluoroethane and two cycles of CsCl-density gradient centrifugation as previously described (21).

For the ELISA-BL, the wells of 96-well, polyvinyl chloride, flat-bottom microtiter plates (Dynatech Laboratories, Inc., Alexandria, Va.) were coated with 100 μ l of a 1:5,000 dilution of hyperimmune guinea pig antiserum to group B ADRV diluted in carbonate-bicarbonate buffer (pH 9.6). The plates were incubated at 4°C overnight, washed twice with PBS containing 0.1% Tween 20 (PBS-Tween), and treated with 1% bovine serum albumin in PBS for 60 min at 37°C. A 25- μ l sample of group B ADRV antigen pretreated with 2 mM EDTA for 10 min was mixed with 50 μ l of twofold dilutions of the serum to be tested and incubated at 37°C for 60 min. The serum samples were diluted to 1:10 in PBS with 0.1% Na₂S₂O₃, and then they were diluted in PBS containing 1% bovine serum albumin. The antigen-serum mixtures were transferred to the wells coated with hyperimmune sera (described above) and incubated for 2 h at 37°C or overnight at 4°C. After five washings with PBS-Tween, 50 μ l of a 1:2,000 dilution of hyperimmune rabbit antiserum to group B ADRV was added, and the plates were incubated for 60 min at 37°C. After an additional five washings with PBS-Tween, 50 μ l of a 1:10,000 dilution of peroxidase-conjugated goat

TABLE 1. Comparison of IEM and ELISA-BL for determining titers of antibody to group A and B rotaviruses

Origin of hyperimmune serum prepared against indicated virus ^a	Group specificity of serum (designation)	Titer of antibody to each rotavirus group detected by indicated test ^b			
		Group A		Group B	
		IEM	ELISA-BL	IEM	ELISA-BL
gp α -SA11	A (BME)	>1,600	10 ⁵	<10	<10
gp α -H-1 ADRV	B (10)	<10	<50	3,200	20,000
la α -H-1 ADRV	B (205)	<10	NT	800	NT
mu α -M-1 ADRV	B (230)	NT	<50	NT	16,000
po α -PRVLA	B (GP 31-4)	<10	<50	320	4,000
bo α -BRVLA	B (H-14)	<10	<50	80	1,000
gp α -ADRV	B (Hung)	<10	<50	160	8,000
mu α -ADRV	B (Hung)	<10	<50	40	200
Rat α -IDIR	B? (Yolken)	<10	<50	40	400
Chicken α -IDIR	B? (Yolken)	<10	<50	20	200
gp α -PaRV	C (12)	<10	<50	<10	<10
po α -PaRV	C (K-619)	<10	<50	<10	<10

^a gp, Guinea pig; la, rabbit; mu, murine; po, porcine; bo, bovine. The specificities of these hyperimmune sera were shown previously (19). PRVLA, Porcine rotaviruslike agent; BRVLA, bovine rotaviruslike agent; IDIR, infectious diarrhea of infant rats; PaRV, pararotavirus.

^b Results indicate the reciprocal of the highest antiserum dilution that showed a positive reaction with SA11 antigen or ADRV antigen.

immunoglobulin G to rabbit immunoglobulin G (Hyclone Laboratory, Logan, Utah) was added. The plates were incubated for 60 min at 37°C and washed five times. ABTS [2,2'-azino-bis(3-ethylbenzthiazolinesulfonic acid)] (100 μ l) in phosphate-citrate buffer (pH 4.0) containing 1.5 μ l of 30% H₂O₂ per ml was added, and the plates were incubated for 30 min at room temperature. The A₄₁₄ was measured with a micro-ELISA reader (Titertek Multiskan; Flow Laboratories, Inc., McLean, Va.).

ELISA-BLs for antibody to group A rotavirus were performed by this same procedure with reagents to group A rotaviruses. In the group A antibody test, the coating serum was a 1:10,000 dilution of hyperimmune guinea pig antiserum to SA11, the antigen was purified simian rotavirus SA11 propagated in monolayers of MA-104 cells as described previously (18), and the detector antibody was a 1:5,000 dilution of hyperimmune rabbit antiserum to group A rabbit rotavirus. For testing for group A antibody, the serum samples were initially diluted 1:50 in PBS containing 1% bovine serum albumin.

In each microtiter plate for the group B ELISA-BLs, an optimal dilution of convalescent-phase serum from a patient with group B ADRV gastroenteritis, which produced about 75% reduction in optical density, served as a positive control; diluent alone served as a negative control. A \geq 50% inhibition (\geq 50% reduction) of optical density produced by the serum samples compared with the buffer control was considered to be positive for antibody to group B ADRV. All tests were performed in duplicate, and the results were averaged.

IEM. Immune electron microscopy (IEM) was performed as previously reported (21).

RESULTS

Development and characteristics of ELISA to detect antibody to group B rotaviruses. Recently, we developed an ELISA to detect group B human or animal rotavirus antigen (21). We have now extended this ELISA to detect antibody to group B rotaviruses by developing an ELISA-BL. Test

TABLE 2. Titers of antibody to group A or B rotaviruses in paired serum samples from Chinese patients with gastroenteritis caused by group B ADRV

Specimen no.	Serum type	Patient		Titer of antibody to ^a :	
		Sex	Age (yr)	Group A rotaviruses	Group B rotaviruses
84-2	Acute phase	Male	37	200	<10
	Convalescent phase			400	160
84-4	Acute phase	Female	29	800	<10
	Convalescent phase			800	160
84-8	Acute phase	Female	51	6,400	<10
	Convalescent phase			6,400	160
84-10	Acute phase	Male	20	400	<10
	Convalescent phase			400	160
84-15	Acute phase	Male	22	1,600	<10
	Convalescent phase			800	160
1	Acute phase	Female	31	3,200	<10
	Convalescent phase			<50 ^b	<10 ^b
2	Acute phase	Female	59	3,200	<10
	Convalescent phase			3,200	80
3	Acute phase	Female	51	800	<10
	Convalescent phase			800	160
4	Acute phase	Male	32	50	<10
	Convalescent phase			50	160

^a Results indicate the reciprocal of the highest dilution of serum that was positive in the ELISA-BL for antibody to group A (SA11) or group B (ADRV) rotavirus.

^b This serum was contaminated with bacteria.

specificity and sensitivity were first evaluated with 10 hyperimmune sera prepared against three groups of rotaviruses (Table 1). The specificities of these sera were previously established by testing by IEM or immunofluorescence with group A rotavirus (SA11), group B rotavirus (ADRV or porcine rotaviruslike agent), and group C rotavirus (porcine paratrotavirus, Cowden strain) (21). A comparison of the titers of antibody in each serum determined by ELISA-BL or IEM is also shown in Table 1. The results of the ELISA-BLs agreed with those obtained by IEM, but the ELISA-BLs were 5 to 10 times more sensitive than IEM. Rat and chicken antisera to the previously unclassified non-group A rat rotavirus also reacted in the group B ELISA-BL for antibody, indicating that this virus may be a group B rotavirus.

These results were obtained with an ELISA-BL in which antigens were premixed with serum samples and the mixture was added to wells precoated with capture antibody (1, 30). By this procedure, all hyperimmune sera to group B rotaviruses and human convalescent-phase sera revealed positive reactions in the ELISA-BLs for group B rotaviruses. The influence of test format was also examined in comparative ELISA-BLs with an alternative format used in blocking radioimmunoassays for Norwalk antibody (14) or human calicivirus antibody (20). In this alternative format, the viral antigens, serum specimens, and detecting antibodies were sequentially added after the plates were coated with convalescent-phase or hyperimmune sera. This latter format showed reduced test sensitivity; hyperimmune sera with high antibody titers by IEM were positive by this sequential procedure, whereas hyperimmune sera with low IEM antibody titers and human convalescent-phase sera that were positive by IEM were negative by this procedure. The same relative test sensitivities were found in the ELISA-BLs for group A rotaviruses. Therefore, we used the mixing procedure to detect antibody in subsequent experiments.

Test specificity and sensitivity were also examined with nine paired serum samples collected from patients with gastroenteritis caused by group B ADRV. These samples

were tested by both group A ELISA-BL and group B ELISA-BL (Table 2). All but one set of paired serum samples (which was contaminated with bacteria) showed a significant (fourfold or greater) rise in antibody to group B ADRV. In contrast, none of these serum samples demonstrated a fourfold or greater rise in titer of antibody to group A rotaviruses. These results were confirmed by IEM, because paired serum samples that were positive for antibody rises with the group B ELISA-BL showed fourfold or greater rises in antibody titer with ADRV antigen but not with group A SA11 antigen (data not shown).

The results of a typical titration of antibody when serum samples from humans or animals were analyzed with the ELISA-BL are shown in Fig. 1. These data also show the observed background (percent reduction) levels seen when negative sera were analyzed in this test. Based on these and other data with negative sera, we used a 50% reduction as the cut-off point to measure antibody in human serum samples from different areas of the world. In addition, because our results indicated that a low proportion of serum samples was positive for antibody to group B rotavirus, any samples that showed positive results with a reduction between 20 and 50% were retested at a 1:2 serum dilution to determine whether antibody was present. All sera that were positive with a 50% reduction always remained positive, and 10 of 11 borderline samples initially showing a 20 to 50% reduction were negative on retesting. The 11 borderline samples were not from a single location.

Prevalence of antibody to group B ADRV. The group B ADRV antibody prevalence in 131 serum samples collected from adults or children without acute gastroenteritis is shown in Table 3. Only 1 sample (from a Chinese staff member who came from the People's Republic of China in 1985) of 19 serum samples collected from staff members working in the laboratory in Houston was positive (titer, 1:20) for antibody to ADRV. Ten serum samples from healthy nonstaff adults, ten samples from adults with Crohn's disease, and ten samples from adults with ulcerative colitis were also negative for antibody to group B rotavirus.

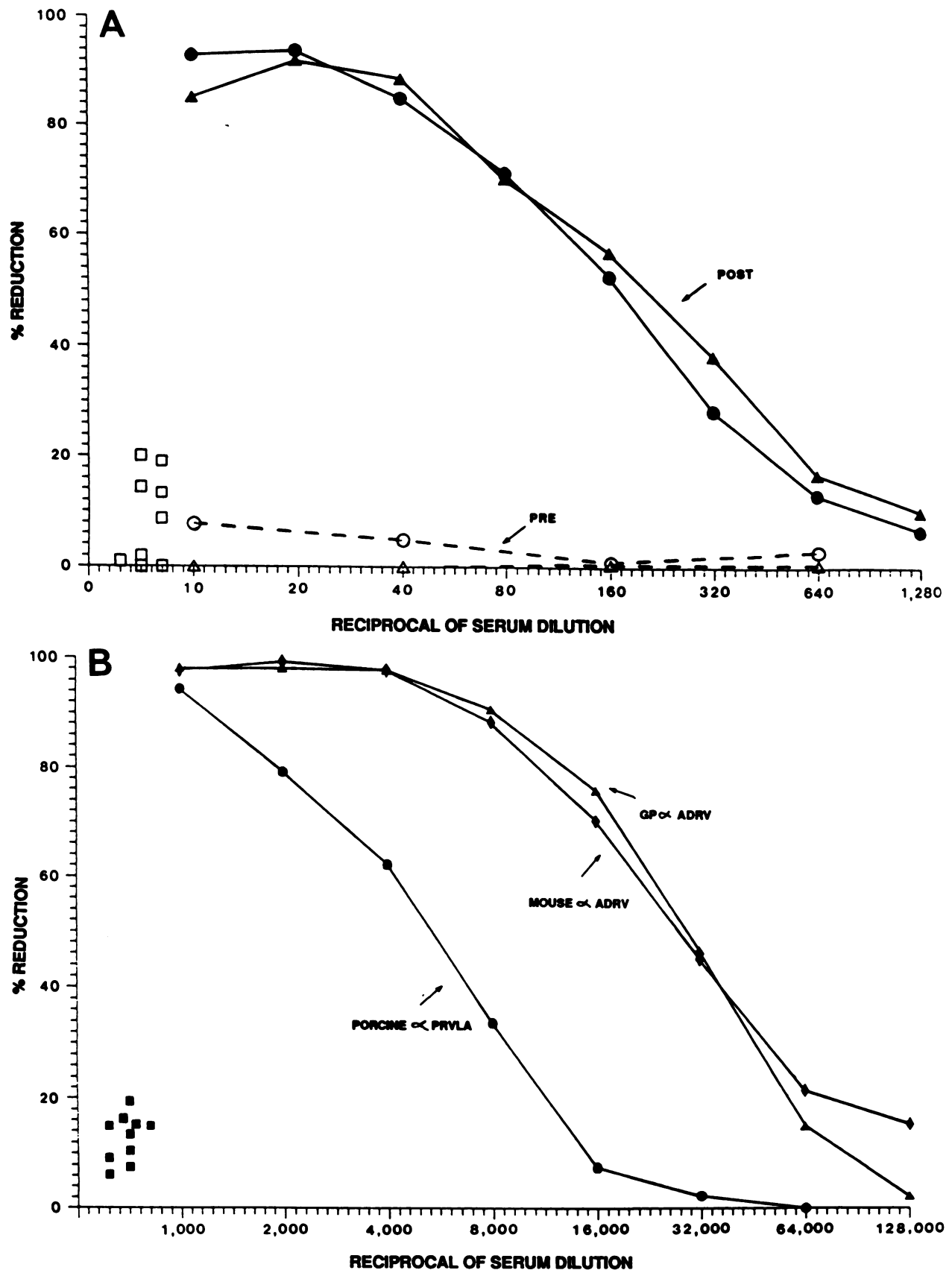


FIG. 1. Titration of serum antibody to group B rotaviruses in (A) paired serum samples obtained from patients with gastroenteritis caused by group B ADRV (Δ , 84-2 [acute phase]; \blacktriangle , 84-2 [convalescent phase]; \circ , 84-15 [acute phase]; \bullet , 84-15 [convalescent phase]) or (B) hyperimmune sera from animals. PRVLA, porcine rotaviruslike agent. A $\geq 50\%$ inhibition of optical density produced by the serum samples compared with the buffer control was considered clearly positive for antibody to group B ADRV. The antibody titers were 1:160 in the convalescent-phase sera and $<1:10$ in the two acute-phase samples. Panel A also shows the results of testing nine other acute-phase serum samples from China (\square). Panel B shows the results of testing six normal guinea pig serum samples, one mouse anti-SA11 serum, one guinea pig anti-SA11 serum, one guinea pig anti-group C porcine paratovirus, and a porcine anti-porcine group C K-619 virus (\blacksquare).

TABLE 3. Prevalence of antibody to group A and B rotaviruses in sera from adults or children without acute gastroenteritis

Serum source ^a	Location	No. tested	No. positive for antibody to:	
			Group A rotaviruses	Group B rotaviruses
Staff members	Houston, Tex.	13 ^b 6 ^c	13 6	0 1
Adults with Crohn's disease	Houston, Tex., and New York, N.Y.	10	10	0
Adults with ulcerative colitis	Houston, Tex., and New York, N.Y.	10	10	0
Healthy nonstaff adults	Houston, Tex.	10	10	0
Hospitalized adults (county hospital)	Houston, Tex.	25	23	0
Texas Children's Hospital patients grouped by age	Houston, Tex.			
0-3 mo		14	14	0
4-11 mo		10	5	0
1-3 yr		10	9	0
4-11 yr		12	12	0
12-19 yr		11	10	0
Total		131	122	1

^a All serum samples were collected after 1984.

^b Three of the staff members were from Hong Kong, Taiwan, or Japan.

^c From staff from the People's Republic of China after 1983.

On the other hand, these 49 sera all had antibodies to group A rotavirus. Most serum samples from 25 adults who were hospitalized in Houston for illnesses other than gastroenteritis also possessed only antibody to group A rotaviruses. We also examined 57 serum samples obtained from children, ages 1 month to 19 years old; none was positive for group B rotavirus, whereas 87.7% of them contained antibodies to group A rotavirus.

The prevalence of antibody to group B and group A rotavirus in 88 additional adult serum samples and 18 immunoglobulin pools from eight countries is shown in Table 4. Only four serum samples (4.5%) were positive for antibody to group B ADRV. The positive serum samples were from Canada (titer, 1:320), Kenya (titer, 1:40), the United States (titer, \geq 1:10), and Thailand (titer, 1:2). None of the immunoglobulin pools from the United Kingdom, Canada, or Japan contained antibody for group B ADRV, whereas all five immunoglobulin pools from China were positive, with low antibody titers (1:40). The titers of antibody to group A rotavirus in the 5 immunoglobulin pools from China and in 3 of the 13 other immunoglobulin pools were between 1:3,200 and 1:6,400. Of these 106 serum and immunoglobulin samples, 104 (98%) were positive for antibody to group A rotavirus.

DISCUSSION

An ELISA-BL specific for antibody to group B rotaviruses was developed and used to evaluate the prevalence of antibodies to group B rotaviruses in serum from eight countries. The results of the serological survey indicate that infections with group B rotaviruses have been much less widespread than infections with group A rotaviruses.

Several possibilities may explain the low prevalence of observed antibody to group B rotaviruses outside of China. First, these rotaviruses may have only recently appeared in China and they may have not yet spread worldwide. The first outbreak of epidemic diarrhea caused by the ADRV oc-

curred in China in November 1982 (17), and since then documented outbreaks involving 19,000 to 20,000 people have been reported only from other parts of China (25, 29). This hypothesis of limited spread is supported by data from China, where the prevalence (41%) of antibody to group B ADRV in regions where epidemics have occurred is reportedly higher than the prevalence (12 to 20%) of antibody in areas where epidemic diarrhea caused by this virus has not appeared (16). Because the clinical manifestations of the illness have been severe (including death of the elderly), outbreaks of diarrhea caused by these rotaviruses are not likely to have been missed if they occurred in other parts of the world, providing virological studies were performed. Our detection of seroconversion (from no antibodies to low titers of antibodies) in the serum samples from people from China who contracted this disease is also consistent with the hypothesis that infections with this virus are a recent event. It would be of interest to examine the prevalence of antibodies to group B ADRV in serum samples collected in China before 1982.

Another explanation for the low prevalence of antibody to group B rotaviruses is that the immune response to these rotaviruses might be different from that to group A rotaviruses. Eight convalescent-phase serum samples from patients infected with group B ADRV revealed low ELISA-BL antibody titers, whereas paired serum samples from the same patients showed higher ELISA-BL titers of antibody to group A rotaviruses. Theil and Saif (26) also reported a low prevalence (23%) of antibody to group B porcine rotaviruses (Ohio strain) in swine sera in the United States by immunofluorescence, and they speculated that either infections with group B rotaviruses fail to induce high serum antibody levels or the group B rotaviruses have a different pattern of infection from that of group A rotaviruses. High levels of antibody to group A rotaviruses are probably maintained in part by repeated clinical or subclinical infections with homologous or heterologous virus strains. The existence of only a few strains of ADRV (compared with the known

TABLE 4. Prevalence of antibody to group A and B rotaviruses in sera from adults from various countries

Serum source	Date collected	No. tested	No. positive for antibody to:	
			Group A rotaviruses	Group B rotaviruses
Kenya	1985	10	10	1
Australia	1981-1983	10	10	0
Thailand	1984-1985	20	20	1
Canada (Ontario)	1983 ^a	5	4	1
	1985 ^a	5	5	0
United States				
Arkansas	1983 ^a	5	5	0
Hawaii	1985 ^a	5 ^b	5	0
Texas	1983 ^a	5	5	0
Maryland	1986	13	12	1
Cruise ship outbreak of gastroenteritis	1985 ^a	5	5	0
Total		88	86	4
Immunoglobulin pools				
United Kingdom	Before 1983	5	5	0
Canada	Before 1983	4	4	0
Japan	Before 1983	4	4	0
People's Republic of China	1986	5	5	5

^a Paired serum samples were analyzed. Acute- and convalescent-phase samples had similar titers for antibody to group B ADRV.

^b These paired serum samples were from a documented outbreak of gastroenteritis caused by Norwalk virus. The convalescent-phase sera all showed a fourfold or greater increase in titer of antibody to Norwalk virus.

heterogeneity in group A rotavirus strains) or limited circulation of these strains, or both, may allow group B antibody titers to decline below detectable levels after infection. Longitudinal studies are needed to address these questions. Interestingly, a high prevalence (86%) of antibody to group B porcine rotavirus (NIRD-1 strain) was found in pig sera collected in 1981 in the United Kingdom (4), but outbreaks or sporadic cases of diarrhea caused by group B rotavirus have not been reported for humans in the United Kingdom. In any event, the low prevalence of antibody to ADRV in human sera suggests that infections (clinical or subclinical) with group B rotaviruses have not occurred in large numbers outside China.

The lack of antibodies in the serum samples from patients with inflammatory bowel disease also suggests that ADRV are not the direct cause of either Crohn's disease or ulcerative colitis. However, our results do not exclude the possibility that any enteric infection might trigger an autoimmune reaction in these patients. These patients have been reported to have elevated titers of serum antibodies to enteric antigens (2), and titers of antibodies to group A and Norwalk viruses were comparable to those of normal controls (15). In any case, the absence of antibodies in these samples provides additional support for the lack of widespread circulation of ADRV outside China.

Reports on the prevalence of antibody to other non-group

A rotaviruses, but not to group B rotaviruses, have revealed a relatively high prevalence of antibodies to non-group A rotaviruses (group C, 77% [4]; group D, 70% [19]; group E, 43% [6]) in pigs or chickens, whereas the reported prevalence of antibody to group C rotaviruses in humans is much lower (17% [10]; 11% [5]). Only one paper has reported a high prevalence of antibody to non-group A rotaviruses in humans (54% in all age groups and 88% in adults [9]). That study used the rat rotaviruslike agent instead of the human group B virus used in the present study as antigen. Although we did not find a high prevalence of antibody in sera from Baltimore, Md., further work is required to understand these different observations.

The present seroepidemiologic studies, together with the surveys described above, suggest that non-group A rotavirus infections are more prevalent in domestic mammals and chickens and that these infections have been uncommon in humans outside of China. It is possible that these viruses are normally animal viruses that occasionally cross species boundaries and infect humans. Biochemical evidence that cocirculating group B ADRV may reassort in nature has been provided by comparative oligonucleotide maps on the genome segments of two strains of ADRV (8). Some serologic evidence for this possibility was provided by the report of the outbreak of gastroenteritis in Baltimore, Md., in children and adults that was apparently caused by a virus similar to the non-group A rat rotavirus (9). Information on this outbreak is needed to clarify the transmission patterns (13). However, the non-group A rat rotavirus should apparently be classified as a group B rotavirus, based on the results of the present study.

Continued surveillance for non-group A rotaviruses, especially for group B rotaviruses, remains important because many people appear to be susceptible to these viruses, and group B human rotavirus infections in China have been extremely pathogenic. Our ELISA and ELISA-BL for group B rotaviruses and for antibodies to these viruses should be useful for this purpose.

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